

Orb prevents autophagy in the *Drosophila* germline through translational repression of *Atg12* mRNA

Isabelle Busseau^{1*}, Stéphanie Pierson¹, Dany Séverac², Christelle Dantec² and Martine Simonelig¹

Supplemental Information

Supplemental Experimental Procedures

Drosophila stocks and genetics

The wild type strain was *w¹¹¹⁸*. The transgenic line used to knock down *Atg12* was #34675 *P{TRiP.HMS01153}attP2* from the Bloomington Stock Center. Other fly stocks were as follows : *orb^{dec}* and *orb^{mel}* (Christerson and McKearin, 1994); *orb^{F343}* (Lantz et al., 1994); *orb³⁶⁻⁵³* (Morris et al., 2003); *twin⁸¹¹⁵* (Zaessinger et al., 2006); *twin^{DG23102}* (Bloomington Stock Center); *nos-Gal4-VP16* (Rorth, 1998); *UASp-Diap1-9-4* and *UASp-Diap1-15a* (Mazzalupo and Cooley, 2006); *FRT82B*, *FRT82B Ubi-GFP* (Bloomington Stock Center); *hs-FLP 122*, *FRT82B orb^{F343}/TM3 Sb*, *FRT82B orb³⁶⁻⁵³/TM3 Sb*. The sequence of *orb^{F343}* and *orb³⁶⁻⁵³* alleles was determined following PCR amplification of exons 3 to 10 using primers indicated below. Flies were raised under standard culture conditions at 25°C. Once collected, females of interest were maintained on medium supplemented with dry yeast until dissection. For the clonal analysis, 2 to 3 day-old females were heat shocked twice daily at 37°C for 1 h with 8 h of recovery, for three consecutive days. Ovaries were dissected 3 to 21 days after the last heat shock. For amino-acid deprivation, flies were transferred to apple juice agar with no yeast for three days before dissection.

Immunostaining

Ovaries were fixed 20 min in PBS containing 4% Paraformaldehyde (PFA), then washed and incubated in PBT (PBS with 0.06% Triton X-100). Incubations were 30 min at room temperature with 0.5 μ g/ml DAPI, overnight at 4°C in primary antibody, and one hour at room temperature in secondary antibody. Primary antibody dilutions were as follows: mouse monoclonal anti-C(3)G 1:500 (Anderson et al., 2005), rabbit anti-Vasa 1:2000 (Lasko and Ashburner, 1990), rabbit anti-Vasa 1:500 (#sc-30210, Santa Cruz Biotechnology), rat anti-Vasa 1:50 (Developmental Studies Hybridoma Bank (DSHB)), mouse anti-Orb 1:5000 (ascites produced from 6H4, DSHB), mouse anti-Hts 1:10 (1B1, DSHB), rabbit anti-cleaved caspase-3 1:500 (Asp175, Cell Signaling Technology), guinea pig anti-Traffic-Jam 1:2000 (Li et al., 2003), mouse anti-Fasciclin 3 1:50 (Fas3, DSHB), rabbit anti-Atg12 1:250 (#OSA00027W, Thermofisher Bioblock), rabbit anti-Atg8 1:1000 (Barth et al., 2011). LysoTracker® Red DND-99 (Invitrogen # L-7528) was used as specified. BrdU incorporation and labeling were performed using the Amersham Cell Proliferation Kit #RPN20 from GE Healthcare. TUNEL staining was done using the ApopTag fluorescein in situ apoptosis detection kit (Millipore #S7110-KIT). Ovaries were mounted in Vectashield and viewed under Confocal Zeiss LSM510 or LSM780 microscopes.

Immunoprecipitations

Ovarian extracts were from 30-50 one day-old females. Precleared ovarian extracts were incubated either in the presence of RNase inhibitor (0.25 U/ μ l RNasin, Promega) or RNase cocktail (0.1 μ g/ μ l RNase A (Sigma) and 0.2 U/ μ l micrococcal nuclease (Sigma) in the presence of 1 mM CaCl₂) with 5 μ l of ascites produced from 6H4 (DSHB, Orb IP) or mouse IgG (Mock IP). Western blots were performed using mouse anti-Orb (6H4) 1:10000 and rabbit anti-CCR4 1:1000 (Temme et al., 2004). RNA extractions were done using Trizol (Invitrogen), followed by RT-qPCR.

RIP-CHIP

Experimental design. For the preparation of the labeled Cy3- and Cy5- aRNA target, seven microliters of RNA from RIPs were amplified and labeled using the Amino Allyl Message Amp II aRNA Amplification Kit (Ambion; Austin, Texas, USA), according to the manufacturer's instructions. Cy3- and Cy5-labeled aRNA samples from Orb RIP and mock RIP were mixed and fragmented with the RNA Fragmentation Reagents (Ambion) to enhance aRNA hybridization, and hybridized on INDAC high-density oligonucleotide microarrays that contained 18,240 spots with long oligomers designed by the International *Drosophila* Array Consortium (<http://www.flychip.org.uk/services/core/FL002/>) representing 14,444 different genes. Prior to hybridization, excess oligonucleotide was removed from the arrays by shaking them twice for 1 min in 0.2% SDS. Arrays were then washed twice in distilled water. The two labeled aRNA were added to version 2 of microarray hybridization buffer (GE Healthcare) in a final 50% formamide concentration, denaturated at 95°C for 3 min and applied to the microarrays in individual chambers of an automated slide processor (GE Healthcare). Hybridization was carried out at 37°C for 12 hours. Hybridized slides were washed at 37°C successively with 1X SSC, 0.2% SDS for 10 min, twice with 0.1 XSSC, 0.2% SDS for 10 min, with 0.1X SSC for 1 min and with isopropanol before air drying. Microarrays were immediately scanned at 10 μ m resolution in both Cy3 and Cy5 channels with a GenePix 4200AL scanner (Molecular Devices) with a variable PMT voltage to obtain maximal signal intensities. ArrayVision software (Alpha Innotech, Santa Clara, USA) was used for feature extraction. Spots with high local background or contamination fluorescence were flagged manually. A local background was calculated for each spot as the median values of the fluorescence intensities of 4 squares surrounding the spot.

Preprocessing. ARRAYVISION software (GE Healthcare) was used for feature extraction. Spots with high local background or contamination fluorescence were flagged manually. A quality analysis was made by generating image plots (MA-plots, boxplot, visualization of the array). No background correction was performed. No spatial bias in the quality analysis was detected. The microarray data were filtered keeping the spot intensity above a median of 90% of the control spots (Empty/Negative Control - NC) plus twice the standard deviation, applied to both channels. A normalization by the median was performed for all microarrays to correct technical bias.

Statistical Analysis. Differential expression was analyzed using the Significance Analysis of Microarrays (SAM) of the siggenes package from Bioconductor (<http://www.bioconductor.org>) within R software (R Core Team 2012; <http://www.R-project.org>). SAM assigns a score to each gene based on the standard deviation of repeated gene expression measurements. Then, a false discovery rate is estimated by permutations of the repeated measurements to obtain a ranking of significantly expressed genes. Multiple testing adjustments were performed by using a false discovery rate approach. The Bioarray Software Environment (BASE) (local installation: <http://baseprod.igf.cnrs.fr/index.phtml>) was used to visualize differential expression for each gene.

Sequences of primers used

RT-qPCR:

Oligonucleotide	Sequence 5'-3'
Atg1 forward 2983	GGATGAGCTACTGAAGAACACG
Atg1 reverse 3127	CAAACGCTTCTCAACGGC
Atg2 forward 59	CACATATATACCCGCCACAG
Atg2 reverse 207	CCACGTTTCGCCTTCTTTG
Atg5 forward 290	ATGGAGCTGTGTGGTTCG
Atg5 reverse 431	ATGTCCTCGGGGAACCTG
Atg7 forward 1996	AAGAGGGTCATGCTTTCCTG
Atg7 reverse 2144	CTAATCCTCGTCGCTATCGG
Atg8a forward 513	TCATTCCACCAACATCGGC
Atg8a reverse 661	TCATTCCGATGCATCCC
Atg12 forward 367	GAACCTGGACCGTAGATCCC

Atg12 reverse 504	TGATTATCTGATCCGGGGC
Atg18A forward 1190	GGAGTGCGCAGGATGTG
Atg18A reverse 1330	ATAGTGGTCCTCCAGCCG
RpL32 forward	CGACGCACTCTGTTGTCTG
RpL32 reverse	CTTCATCCGCCACCAGTC

PAT assays:

Oligonucleotide	Sequence 5'-3'
Atg12 forward 350	GCCCATCATCAAAAAGCGAACCTGG
RpL32 PAT	CTGCCCACCGGATTCAAGAAGT
Arpc2 PAT	CTCCGCCAATTTGTTACAGTCATAG
aret PAT2	TAGTTTCTATTATTTGCTACATCATCC
CCT2 PAT	GTCCCTGTCGTGTAATCCTTCTAC
cup PAT	GAGTCAAGACCCAAACGGAGCG
CycB PAT	GCTGGCCGAACACATCGGCG
dhd PAT	GTAAGCGCGAGATGTGGGTAGC
Dph5 PAT	GCACTCGCTCATCATTCCCGCC
gnu PAT	CCGATTGGCCATGAATATTATCC
His1 PAT	CATAAAGTCGAAGCCCGCCG
His2A PAT2	TATTCGCGTCATCTGCAACTGGC
His2B PAT	GGCGATGAGCATAATGAACAGC
His3 PAT	CGCTCAGGACTTTAAGACGGACTTG
His4 PAT	GTGCTGCGTGATAACATCCAAGGTA
me31B PAT	ACCCCGCCCTTTATGTGCGAAACG
mtrm PAT	GACCTGGTGTCTTAAAGCTCGTCG
mus209 PAT	GTCGATGTGCGCAGATGTTCC
osk PAT2	AAGCGCTTGTTTGTAGCACA
QC PAT	GCATTTGGTAGCCACTCCTTTCCC
SmG PAT	CGAAGAACAACATCGGCATGGTGG
smt3 PAT	GTTACTCCTCTTACAACACTACACACTT

RNA pull-down assays:

To generate a fragment of 326 bp containing *thread* (*th*) 3'UTR:

Oligonucleotide	Sequence 5'-3'
th T7forward 1513	TAATACGACTCACTATAGGGAAGCCCTTCACCGATGTGATG
th reverse 3'flank	CTGCCACGAGTGCCATGAGTTTTTC

To generate a fragment of 393 bp containing *Atg12* 3'UTR:

Oligonucleotide	Sequence 5'-3'
Atg12 T7forward 350	TAATACGACTCACTATAGGGCCCATCATCAAAAAGCGAACCTGG
Atg12 reverse 3'flank	GTGTGAAATCGACTTCGCAACATCATC

To generate a fragment of 1096 bp containing *osk* 3'UTR:

Oligonucleotide	Sequence 5'-3'
osk T7forward 1806	TAATACGACTCACTATAGGGGCAAGTTATTGAAACGAGTCTGG

osk reverse 3'flank	GAACATAGCTTAGAGCAAACAAAATCATTG
---------------------	--------------------------------

To generate fragments of 1670 bp without CPE, or of 1788 bp with CPEs from *pTRI-Xef* template (Ambion MEGAscript kit):

Oligonucleotide	Sequence 5'-3'
TRI forward	GATTTAGGTGACACTATAGAATACACGG
TRI reverse no CPE	TCTGAAGCTCTTGCGATGCATTG
TRI reverse CPE	GGTCTCAAATTTGGTGACAGATTTTGG

Genomic PCR and sequencing of *orb*:

Oligonucleotide	Sequence 5'-3'	Sequenced <i>orb</i> region
orb forward exon3	AAGTTTCAGCAACTGTTCGACCG	Exon 3
orb reverse exon3	ACAAAGTAAAAGAACAACCCACTG	
orb forward exon4	TGAGGGCTGGACAAACTGTGC	Exon 4
orb reverse exon4	GAGTGCAGTTAGAGAGAAGTTGAAG	
orb forward exon5	GAAGTTACCTATTGTCTGTTACCGT	Exons 5-6
orb reverse exon6	GTGAACGAAAAATCTATGGTGTC	
orb forward exon7	AGTTTATCCTCTAAATCATTCGTCCT	Exon 7
orb reverse exon7	TTGCCGGGCCACTCCACTC	
orb forward 2095	GTGTGGGTGCTTCTAGTAGCGG	Exon 8
orb reverse exon8	GTTTCTGGGAGCTGGATCGCAC	
orb forward 2744	CTCTTCGCGGCGTATTAAGTCCAAG	Exon 9
orb reverse exon9	GAGTATTCTTGACAGCTGACGGTG	
orb forward exon10	GTTTCGTTACCTTCTCTGGCC	Exon 10 (coding part)
orb reverse 3923	ACTTGAGTAGTATTTGTATGAGGGGT	

References

- Anderson, L.K., Royer, S.M., Page, S.L., McKim, K.S., Lai, A., Lilly, M.A., and Hawley, R.S. (2005). Juxtaposition of C(2)M and the transverse filament protein C(3)G within the central region of Drosophila synaptonemal complex. *Proc Natl Acad Sci U S A* 102, 4482-4487.
- Barth, J.M., Szabad, J., Hafen, E., and Kohler, K. (2011). Autophagy in Drosophila ovaries is induced by starvation and is required for oogenesis. *Cell Death Differ* 18, 915-924.
- Christerson, L.B., and McKearin, D.M. (1994). *orb* is required for anteroposterior and dorsoventral patterning during Drosophila oogenesis. *Genes Dev* 8, 614-628.
- Lantz, V., Chang, J.S., Horabin, J.I., Bopp, D., and Schedl, P. (1994). The Drosophila *orb* RNA-binding protein is required for the formation of the egg chamber and establishment of polarity. *Genes Dev* 8, 598-613.
- Lasko, P.F., and Ashburner, M. (1990). Posterior localization of vasa protein correlates with, but is not sufficient for, pole cell development. *Genes Dev* 4, 905-921.
- Li, M.A., Alls, J.D., Avancini, R.M., Koo, K., and Godt, D. (2003). The large Maf factor Traffic Jam controls gonad morphogenesis in Drosophila. *Nat Cell Biol* 5, 994-1000.
- Mazzalupo, S., and Cooley, L. (2006). Illuminating the role of caspases during Drosophila oogenesis. *Cell Death Differ* 13, 1950-1959.

Morris, J.Z., Navarro, C., and Lehmann, R. (2003). Identification and analysis of mutations in bob, Doa and eight new genes required for oocyte specification and development in *Drosophila melanogaster*. *Genetics* 164, 1435-1446.

Pique, M., Lopez, J.M., Foissac, S., Guigo, R., and Mendez, R. (2008). A combinatorial code for CPE-mediated translational control. *Cell* 132, 434-448.

Rorth, P. (1998). Gal4 in the *Drosophila* female germline. *Mech Dev* 78, 113-118.

Temme, C., Zaessinger, S., Meyer, S., Simonelig, M., and Wahle, E. (2004). A complex containing the CCR4 and CAF1 proteins is involved in mRNA deadenylation in *Drosophila*. *Embo J* 23, 2862-2871.

Xie, T., and Spradling, A.C. (1998). decapentaplegic is essential for the maintenance and division of germline stem cells in the *Drosophila* ovary. *Cell* 94, 251-260.

Zaessinger, S., Busseau, I., and Simonelig, M. (2006). Oskar allows nanos mRNA translation in *Drosophila* embryos by preventing its deadenylation by Smaug/CCR4. *Development* 133, 4573-4583.

Supplemental Figure legends

Supplemental Figure 1: *orb* null mutations do not affect germ line stem cells

(A) Molecular organization of the *orb* locus, showing the *orb-RA* mRNA (Flybase) and alterations in *orb* null mutants. *orb^{dec}* has a *P*-element inserted in the second exon, and sequencing of *orb^{F343}* and *orb³⁶⁻⁵³* revealed non-sense mutations creating premature stop codons.

(B) Predicted full-length and truncated Orb proteins from wild type, *orb^{F343}* and *orb³⁶⁻⁵³* alleles. Grey boxes indicate glutamine-rich domains, black boxes indicate RNA recognition motifs (RRM).

(C) Expression of Orb in early germ cells. Germarium stained with anti-Orb (white/red), anti-Bag-of-marble (Bam) (green) and DAPI (blue). Orb expression alone is shown in the middle panel; the left panel is an enhanced view of Orb expression showing low levels of Orb in the GSCs (yellow arrowhead). Orb was present at very low levels in the GSCs and mitotic germ cells, and its expression increased in region 2a of the germarium, in 16-cell cysts.

(D-F) Clonal analysis of *orb^{F343}*. (D) Germaria were marked with GFP (green), 1B1 antibody which labels spectrosomes and fusomes (white) and anti-Vasa (red). *orb^{F343}* mutant GSCs (dotted yellow outline) were able to differentiate into cysts marked with fusomes (continuous yellow outline). (E) Loss of wild-type and *orb* mutant clonal GSCs occurred at similar rates indicating that *orb* is not required for GSC maintenance. Quantifications were performed as in (Xie and Spradling, 1998). (F) Division rate of *orb* mutant GSCs. The ratio of [GFP-, clonal] over [GFP+] cysts, in germaria containing one [GFP-] and one [GFP+] GSCs, was calculated and found to be close to 1 for both the wild-type and *orb* mutants, indicating that *orb* is not required for GSC division.

Supplemental Figure 2: Experimental design of the RIP-Chip experiments

Supplemental Figure 3: Sequences of Autophagy-specific gene 3'UTRs containing CPEs

Motifs identified using the software designed in (Pique et al., 2008) are indicated. CPENC, predicted non canonical cytoplasmic polyadenylation element; CPEC, predicted canonical cytoplasmic polyadenylation element; PBE, predicted Pumilio Binding Element; HEXA, predicted hexanucleotide of canonical polyadenylation signal.

Supplemental Figure 4: Starvation-induced cell death is sensitive to *orb* gene dosage

(A) DAPI and TUNEL staining of ovarioles arrested before vitellogenesis in wild-type females after three days of amino-acid deprivation. White arrowheads indicate degenerating egg chambers.

(B, C) Quantifications of ovarioles showing mid-oogenesis arrest in wild-type (w^{1118}) or heterozygous orb^{F343} and orb^{dec} females raised on rich food (B) or after three days of amino-acid deprivation (C). **: $P < 0.01$, ***: $P < 0.001$, using the χ^2 test.

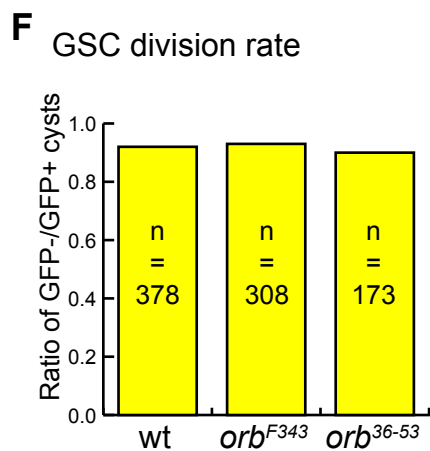
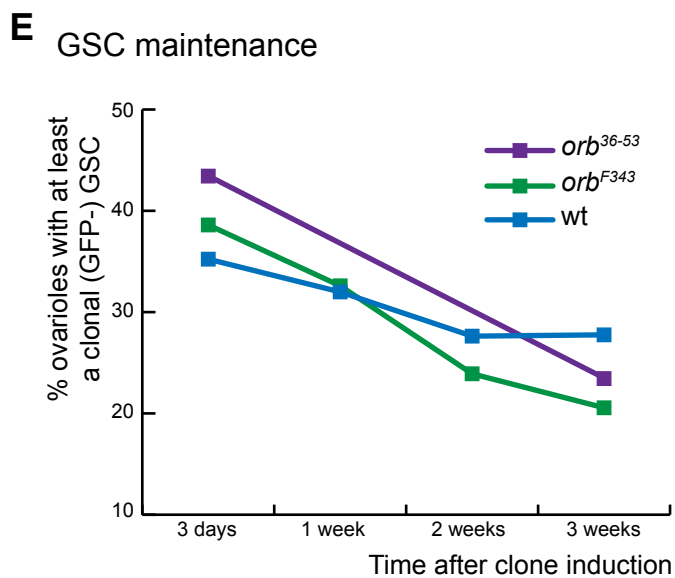
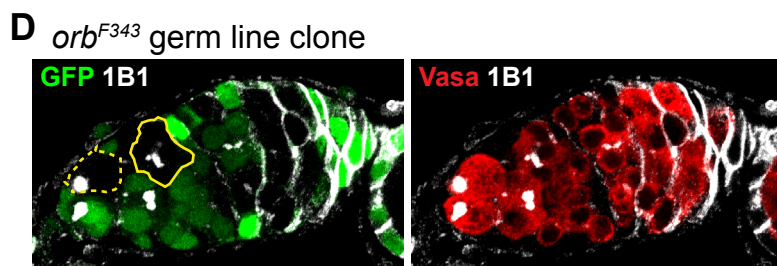
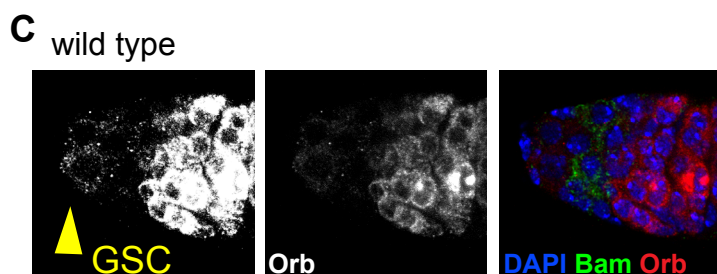


Figure S1

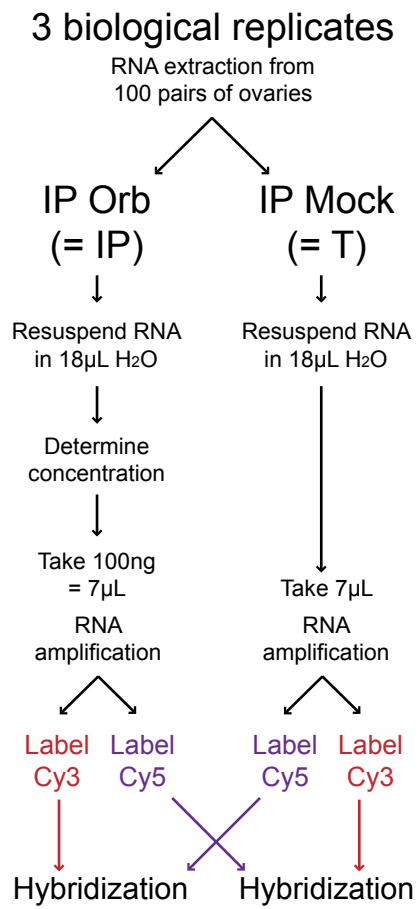


Figure S2

3'UTR *Atg1-RA*

TAGTTACTCGGGCGGATAGAGCTATATTCCAACCATGAAGCACCGGCAATCTTCACAACATCAACAACATTTCTTAAGTTAT
TCACCGTTTTCTGTTGGGAGTGCAAACATGTGATTGAAACAAAAATACACTATCCATACATAGACATACCTACAAATAATTTA
ACAAACCATTCACACCTAATTTCTGGCCTTCAAAGTTTCCCAACAAAAAAGAAAAAACAGTCCAA**GTGAAGT**TTTG
AGCTATGCAGATGTATGTAGTTTTTGTCTATATAATTTGGCCCTTGCGGCAACCATAATTGTTAAAGCTTATAACGAG
TGCAGATGCAACATGAATAAGGGCAGACAAGAGTCCATATAGTCGTAAGATAACCATAATATTATATATATGTTTACATATAT
GTACAACACGAAAGCTAACCAAGCAAACACATTACATAACGTAAAGGGCAATATGCAAGCCAAACTCAGTTTAAATCTATGT
TTAAAGGAGGAAGCGGGAGAAGCCAGATCCATATATCATGTGCTAGAATTTCTCCGTAG**TTTAA**TGCCTATTTTCTTATTTA
ACAATCTAATATTTCCCTATTATTATATTATAAACATACCATAACAATATTGATGGCTGCACTACACAACATGTTTACAATGT
GCTATGTTTCATAGTTTTTAGTTAGGTCCTTTCTAAAACAACCTTTAAAGATGTATTTAA**TTTAT**GACAAGAAATTTATATTTT
AAAGAAACT**TTTAA**GTTTTCTACAAGCAAAGCCACAAAACCTTCGAAAAATCTATATATGTATATGGTATACAAAAGAG
AGAAACACATAAAAGAGTGTATAAAATGAAAAAGAAACAAAATACCGATTTGAAC**ATAAA**AGTACTAAGGAAATCGT
Motifs found: #CPENC=1 #PBE=1 #CPEC=2 #HEXA=1

3'UTR *Atg2-RA*

TAGGCAACTGCTTCACAGATCGACTAGGTTAACGGTAGCTATAGTTCACGGGGCTTACAGCATAACGCAATGTTTTCTATTGT**TTTACT**AAT**ATAAA**TAAT**GTACATA**AAAAGGAGCACTTGCACGAGACGGCGAGGTGACGAGGGATTGGGATCCCGAACGGTG
ATTCGACATGGGTGTGATTGAACCTTATCCTTTTTAGCCATATTACAGTACATGAGAGTTATATTT**TTTTAT**TATTTTACCTG**ATAAA**TATATATATATATAAAAGAAAAAG
Motifs found: #CPENC=1 #PBE=1 #CPEC=1 #HEXA=2

3'UTR *Atg5-RA*

TAGCCTTATCCCTATAT**GTATATA**GCCCTATGTTTTGCCAGTTCGCGTGCCTGTATATTATGTTTGTGTTTCGTGTCAATGT
GTGCTTGTGAGAGTGTGTGCAGTTCTATTTTTCGATTCCAGCAAATCCAGGGCGATCCTCTGCGATCGATTTACATACATAT
ATACGGATCCAT**GTATATA**TCCAAATATATGTCTAGTTAAGCTAATCTGTATGCTATATTATGATATATGCCAGCATTTAAA
AGAAGCGAGCAGCGCAAACCAAACGATTAATGAAACTACCCACTAATAATAATGCCACATACAGTTAACTTAACTTTTTT
TTTTTTTTTGGATCATAATCTGCCGACTTTAGTTGCGAAAACTTGATTGAA**ATTAA**GCACAG**TTTTAT**TACGTTTATGTAA
TTATTGTATACACTAAATGACATCACGCCACACTAATGATTAATGGAACTAT**TTTTAT**ATCGACCCCAATCAAAAATATATAT
ATATACGAAAGTTAT
Motifs found: #CPENC=0 #PBE=2 #CPEC=2 #HEXA=1

3'UTR *Atg7-RA*

TAGGTTCCACAAATTTGGTCAAG**TTTTAT**CTTCAAAGCAAAATATCTTAAATATGAAATTCAGCTTATTGTGGCTTTAATTTT
CTGCAGCATTTATAGTTCAATGTCTATACCAAAATGTGTGTACTTCAATGC**TTTTAT**TCTATCTAGTCGTTAAGCAAATGT
GAATCATTTCAAATGAAATTTGATGTGAAATTAGGCATAAGAAAAATGTGAATTTACTATTGAAATTTGTAAT**ATAAA**AGTGT
GAATTTACTATTGAAATTT
Motifs found: #CPENC=0 #PBE=0 #CPEC=2 #HEXA=1

3'UTR *Atg8a-RA*

TAACTTTGCTCCGGTCGGGATGCATCGGAATGAAGCCCCCCCCCTATACAGAAATCTATATATA**ATAAA**ATATATTTTGTGT
TTAGCATGCAAGCTTACAATATGGACGGCGAGTTAGCTAAACGAATTGAGGAAAACCGCAGGAAATGCCACATGAAAAGAACT
TGAAATTGATCTGATACGCAGGGCAACAAAAAAATCCACAAAACCGAAAACTACAAAAACAATTGTTATGTGAACATTAT**TT**
TTATATATATACATTTTCAAACGCGTTAGTATTTCAA**TTAA**CTCTGCTTTGTGTCTACCCATTACGAAACAACCAACCA
ACCAACTTTCCCCACAAAAACAGATCTATTACACCCCTTCTGGGCCATTTAATTTCCATTGCTGTAATTGATCCGTGCGAA
TGCTTTGTACTTTACTTTGTACTTTGCTAATATTTCAATGAATTCATATTTTTTT**TTTAT**TCACGATGCAACAAATTTCT
GTGCATATTTAGTGTATGTTACGAATAGGACTCTCTCGGTAAAGTGTACGGTAATTAATCTATT**ATTAATAAA**CAAAAA
CC
Motifs found: #CPENC=0 #PBE=0 #CPEC=2 #HEXA=4

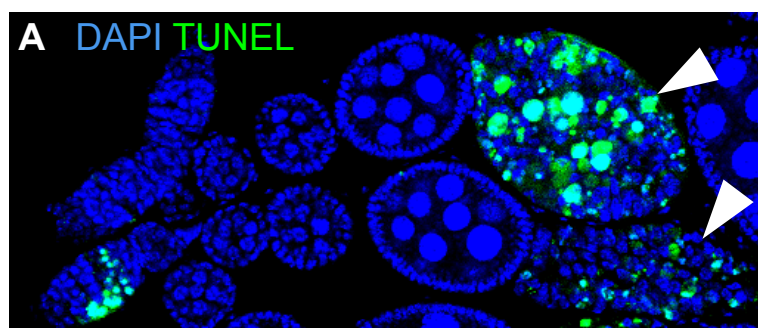
3'UTR *Atg12-RB*

TAAATCGATACGCACATGACTTTGCTAAGTCTTAAGTA**TTTTAT**TCGACTACTGTGTAAT**TTTTAT**ATT**ATAAA**TGTGAATAA
GATG
Motifs found: #CPENC=0 #PBE=0 #CPEC=2 #HEXA=1

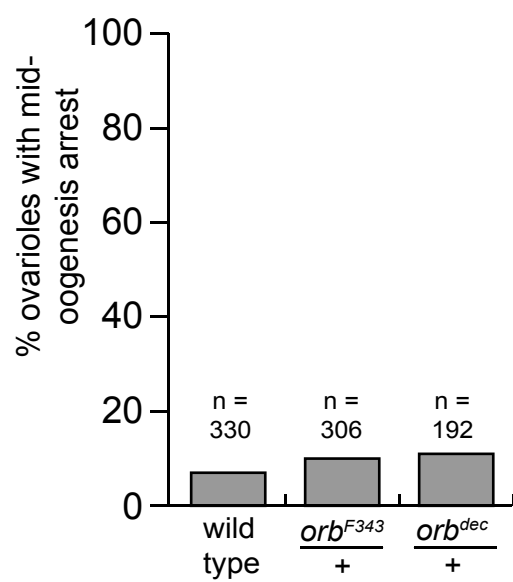
3'UTR *Atg18-RA*

TAACTGTTTCGCTTCTCGCACACACTTCACACTTACACACATCCCAACCAAACCCACAATCGAAACAAATTTCTCTAGTTGATTC
ACCTCAAACCTCTAGGCCTTAACAGTGGCGTGAGCATTTGGCGGAGCAGCTGGAGTTCCGCCGAGCAGTGCAAGTGCAGCCGGAG
CAGGCATGGTGGAAGTGGCGGTGATGGCAAGTCGGCTGTTTCTGTTACCGGTGGTGTGACAAAACCAAGTAGCTCCTATGCG
TCGGCCGTGAAGGGTGACGATCCCGTGGGACCCTCATCCGTTAGACAAGGTCAATGATCAAAAATGAGAGATGCAGCGATGG
TAAACGAAACTCACTCCTCAAAGTAATAAGGACGATGATGCTAAAGTGATTACAATGATGCTTGGCGACTGGCGGCGCGGATA
ATGATGATG**ATAAA**CGACGGTGGCTCTGTGGCGTCAAGCGTATTTAAGTA**ATAAA**TAGGTCATAGACTTAATTTAGCTCTT
AACGTTGTGAGCGATGATTCTTCCTCGTTTTGTACCAGCCAATATTCCTTTGTACGGCTCTCCCTACCCAAAACATTTTTTA
GTTTTAAGCCTCCGTTCTGCGCTGCCATTGGAATGTGAAATGCTTTTGTATACACTCCTAAGTTAGTTGTTAATTAATAA
TTTAGACGGATTTACAGATTCGCTTTGATATTTGAGTTGCCAGCT**TTTTAT**ACAA**ATAAA**CGGCCCTTTGCAAATT
Motifs found: #CPENC=0 #PBE=0 #CPEC=1 #HEXA=3

Figure S3



B rich diet



C poor diet for 3 days

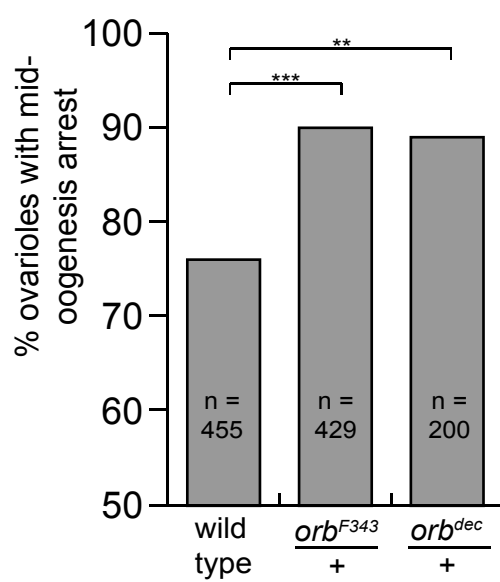


Figure S4

Table S1: mRNAs present in Orb RIP. Gene ID and fold changes (intensity in Orb RIP/intensity in Mock RIP) ≥ 1.5 using SAM with FDR ≤ 0.01 are indicated.

FlyBaseID	Symbol	Fold change mature ovaries	Fold change early ovaries	Presence of CPE
FBgn0011710	sep1	1.81		
FBgn0266375	IP3K2	4.1		
FBgn0010339	128up	1.7		CPE
FBgn0020238	14-3-3epsilon	1.82		
FBgn0038363	Acyp2	1.94	2.67	
FBgn0037555	Ada2b	1.83		CPE
FBgn0027493	AdSS	3.27		
FBgn0014455	Ahcy13	2.81		
FBgn0027932	Akap200	1.61		
FBgn0020764	Alas	1.69		
FBgn0260972	alc	1.53		
FBgn0000064	Ald		2.11	CPE
FBgn0012036	Aldh	2.96	2.89	
FBgn0003884	alphaTub84B	14.01	5.34	
FBgn0003885	alphaTub84D	3.3	3.24	
FBgn0029512	Aos1	4.11	3.36	
FBgn0034231	APC10	1.59	2.06	CPE
FBgn0038742	Arc42	1.82	2.37	CPE
FBgn0000114	aret	3.72	3.06	CPE
FBgn0011745	Arp1	1.5		
FBgn0011741	Arp6		1.87	
FBgn0032859	Arpc2		2.15	
FBgn0031781	Arpc4	2.86	2.65	
FBgn0031437	Arpc5	1.87	2.33	CPE
FBgn0036826	arx	1.68		
FBgn0036255	Atg12		1.9	CPE
FBgn0019644	ATPsyn-b	1.67		
FBgn0010217	ATPsyn-beta	1.62		
FBgn0016120	ATPsyn-d	1.95	2.55	
FBgn0020235	ATPsyn-gamma	7.66	4.1	
FBgn0004587	B52	4.26		
FBgn0045866	bai	1.65		
FBgn0260960	Baldspot		1.7	
FBgn0027889	ball	1.56		
FBgn0000173	ben	4.11	2.73	CPE
FBgn0010357	betaTry	2.47	3.03	
FBgn0003887	betaTub56D	7.61	4.01	
FBgn0024491	Bin1	2.85	2.37	CPE
FBgn0010300	brat	2.13	2.52	CPE
FBgn0032679	bsf	1.62	2.17	
FBgn0031696	Bub1	2.17		
FBgn0020556	bxd		2.77	
FBgn0000250	cact	2.95		CPE
FBgn0015614	CanB2	1.76		
FBgn0022943	Cbp20	2.86		
FBgn0261353	Ccdc56	1.7		

FlyBaseID	Symbol	Fold change mature ovaries	Fold change early ovaries	Presence of CPE
FBgn0030086	CCT2	5.5		
FBgn0034443	cer	1.77		
FBgn0039112	CG10219	2.09	2.34	
FBgn0022344	CG10340		2.01	CPE
FBgn0032724	CG10428	1.94	1.83	
FBgn0033017	CG10465	3.34	2.39	CPE
FBgn0034583	CG10527	1.96		CPE
FBgn0035630	CG10576	3.51		
FBgn0035603	CG10635		2.25	
FBgn0035592	CG10674		2.42	
FBgn0030518	CG11134	2.08	2.31	
FBgn0037199	CG11137	6.86	3.33	
FBgn0036334	CG11267	1.61		CPE
FBgn0036342	CG11279	2.31		
FBgn0039868	CG11563		2.1	CPE
FBgn0030545	CG11590		2.61	
FBgn0030551	CG11674	3.43		
FBgn0030292	CG11752	3.33	2.85	
FBgn0039259	CG11781	2.03		
FBgn0039265	CG11790	1.82		
FBgn0039305	CG11858	2.19	5.08	CPE
FBgn0031253	CG11885		2.34	
FBgn0037652	CG11980	1.57	2.02	
FBgn0040534	CG11985	1.76		CPE
FBgn0035404	CG12079	1.75		
FBgn0033473	CG12128	1.6	2.14	CPE
FBgn0031048	CG12237	1.5		
FBgn0035811	CG12262	1.63		
FBgn0038080	CG12279	1.94		
FBgn0032620	CG12288	2.14	2.52	
FBgn0033624	CG12384	2.32		CPE
FBgn0037368	CG1239		1.93	
FBgn0035370	CG1240	2.09		
FBgn0031505	CG12400	3.34	3.62	
FBgn0036872	CG12519	13.25	8.5	
FBgn0037779	CG12811		2.09	
FBgn0040666	CG12848	2.56	2.63	
FBgn0033961	CG12859	7.83	4.72	
FBgn0033507	CG12909	3.19	2.67	
FBgn0037061	CG12975	2.13		
FBgn0040751	CG13018	2.06	2.06	
FBgn0036665	CG13024		1.82	
FBgn0036599	CG13044	1.63		
FBgn0036594	CG13047	6.66	3.51	
FBgn0036607	CG13059	3.95	2.69	
FBgn0026566	CG1307	3.44		
FBgn0032127	CG13114	1.55		
FBgn0033608	CG13220	2.61		
FBgn0035694	CG13299	2.84		
FBgn0033850	CG13331	1.77		

FlyBaseID	Symbol	Fold change mature ovaries	Fold change early ovaries	Presence of CPE
FBgn0026879	CG13364	2.26	2.98	CPE
FBgn0032033	CG13392	1.56		
FBgn0040660	CG13551	3.98	2.59	
FBgn0035858	CG13674	1.83		CPE
FBgn0033340	CG13751	1.76	2.7	
FBgn0040954	CG13779	1.56		
FBgn0035323	CG13807		1.69	
FBgn0031776	CG13993	3.91	2.46	
FBgn0040949	CG13998		1.69	
FBgn0031677	CG14036	4.14		
FBgn0036871	CG14096	1.8		
FBgn0030584	CG14407	1.5		CPE
FBgn0033000	CG14464	1.75		
FBgn0034245	CG14482	2.93	3.91	
FBgn0034248	CG14483		2.01	
FBgn0031943	CG14538		1.77	
FBgn0039404	CG14543	2.18	2.85	
FBgn0040602	CG14545	5.44	2.68	CPE
FBgn0037340	CG14671	2.02	2.06	
FBgn0037883	CG14701		2.07	CPE
FBgn0029594	CG14806	2.47	2.47	
FBgn0026090	CG14812	1.65		
FBgn0026089	CG14817		2.58	
FBgn0038428	CG14894	1.73		
FBgn0038437	CG14898		1.98	
FBgn0035415	CG14966	2.01	2.28	
FBgn0035528	CG15012	1.6	2.05	
FBgn0035541	CG15019	1.66	2.47	
FBgn0030174	CG15312	1.58		
FBgn0031143	CG1532	1.61		
FBgn0030104	CG15368		1.75	
FBgn0040650	CG15456	1.59		
FBgn0033225	CG1550	1.89		
FBgn0039732	CG15525	1.87		
FBgn0036909	CG15881	3.38	2.76	
FBgn0030704	CG15916	1.55		
FBgn0040575	CG15922	1.7	2.04	
FBgn0033183	CG1620	1.7		
FBgn0037728	CG16817	2.16		
FBgn0040754	CG17059		2.47	
FBgn0038107	CG17327	1.71	2.03	
FBgn0030239	CG17333		2.23	
FBgn0039830	CG1746	10.12	6.18	
FBgn0083978	CG17672	25.66	5.29	
FBgn0039993	CG17691	2.33		CPE
FBgn0037885	CG17721	2.58		CPE
FBgn0035423	CG17737	5.14	4.72	
FBgn0035425	CG17746	1.6		
FBgn0040899	CG17776	10.69	5.55	
FBgn0023537	CG17896	1.67		

FlyBaseID	Symbol	Fold change mature ovaries	Fold change early ovaries	Presence of CPE
FBgn0036537	CG18081	5.06		
FBgn0029971	CG18624	2.02		
FBgn0039869	CG1890	1.8		
FBgn0039690	CG1969	10.17		CPE
FBgn0035271	CG2021	1.52		
FBgn0000299	Cg25C		2.59	CPE
FBgn0031263	CG2789	2.8		
FBgn0034753	CG2852	3.78		
FBgn0031459	CG2862	3.4	2.54	
FBgn0030170	CG2990		2.76	
FBgn0050109	CG30109	1.74		
FBgn0050122	CG30122	1.82		CPE
FBgn0050159	CG30159		1.86	
FBgn0050185	CG30185	1.58		
FBgn0050349	CG30349	1.5		
FBgn0050382	CG30382	2.83		
FBgn0250838	CG30415		4.14	
FBgn0050423	CG30423	3.03		
FBgn0050499	CG30499	3.49	3.01	
FBgn0051109	CG31109	1.87		CPE
FBgn0051279	CG31279		1.5	CPE
FBgn0051472	CG31472	1.53		CPE
FBgn0051548	CG31548	1.62	2.44	
FBgn0051638	CG31638		1.71	
FBgn0051715	CG31715	2.3	2.76	
FBgn0051957	CG31957	1.88	1.95	
FBgn0052068	CG32068	2.01		
FBgn0052069	CG32069	1.64		
FBgn0031436	CG3214	1.86	2.67	
FBgn0052230	CG32230	3.09	4.54	
FBgn0029882	CG3226	2.33		
FBgn0047135	CG32276	3.22		
FBgn0052278	CG32278	1.95	2.71	
FBgn0052344	CG32344		2.04	
FBgn0052428	CG32428	1.75		
FBgn0052448	CG32448		2.63	
FBgn0052500	CG32500	1.57		
FBgn0052736	CG32736	1.76	2.32	
FBgn0052797	CG32797	1.73		
FBgn0053155	CG33155	4.15	2.75	
FBgn0053170	CG33170	1.57		CPE
FBgn0038224	CG3321	25.15	10.32	
FBgn0053229	CG33229		2.33	
FBgn0053493	CG33493	2.24		
FBgn0053502	CG33502	1.57		
FBgn0064116	CG33713	1.68		
FBgn0064117	CG33714	1.68		
FBgn0085405	CG34376	8.39	5.38	CPE
FBgn0029868	CG3446	1.77	2.35	
FBgn0030733	CG3560	5.6	3.68	

FlyBaseID	Symbol	Fold change mature ovaries	Fold change early ovaries	Presence of CPE
FBgn0029854	CG3566	3.31		
FBgn0023521	CG3587		1.88	
FBgn0025839	CG3621	3.47	3.39	
FBgn0031600	CG3652	1.79		
FBgn0035044	CG3663	1.5	2.51	
FBgn0035987	CG3689	2.31		
FBgn0040346	CG3704	1.58		
FBgn0038271	CG3731	1.99	2.29	
FBgn0022343	CG3760	2.94	2.65	
FBgn0029867	CG3847	1.87		
FBgn0036824	CG3902	2.71		
FBgn0040396	CG3939	5.31	5.24	
FBgn0058042	CG40042	7.72	4.29	
FBgn0262116	CG40127	4.87	3.93	
FBgn0058315	CG40315		1.54	
FBgn0025558	CG4101	1.65		
FBgn0250814	CG4169	4.09	2.58	
FBgn0259223	CG42323	4.88	3.78	
FBgn0259238	CG42336	3.53	3.17	
FBgn0259704	CG42358		1.88	
FBgn0259705	CG42359		1.88	
FBgn0260767	CG42565	1.7		
FBgn0260768	CG42566	1.7		
FBgn0038313	CG4338		1.97	
FBgn0032138	CG4364	2.21	1.81	
FBgn0265177	CG44242	2.18	1.91	
FBgn0265178	CG44243	2.18	1.91	CPE
FBgn0030792	CG4789	2.17	2.09	
FBgn0039563	CG4951	1.78	2.26	
FBgn0032217	CG4972	1.65		
FBgn0030817	CG4991	3.77		
FBgn0260747	CG5010	2.46		
FBgn0030830	CG5172		1.62	CPE
FBgn0013763	CG5210	1.76		
FBgn0037891	CG5214	1.84	2.26	
FBgn0034362	CG5323	1.75		
FBgn0032216	CG5384		2.02	
FBgn0030605	CG5548		2.66	
FBgn0039537	CG5590	2.25	2.12	
FBgn0032190	CG5739	1.83		
FBgn0038858	CG5793	1.69	2.6	
FBgn0035926	CG5804		1.58	
FBgn0015338	CG5861	1.79		
FBgn0027586	CG5867		2.00	
FBgn0028894	CG5869		1.82	CPE
FBgn0036991	CG5872		3.01	
FBgn0039490	CG5882		1.74	
FBgn0036998	CG5969	1.87		
FBgn0037001	CG6020	2.35	2.83	
FBgn0040985	CG6115		1.58	

FlyBaseID	Symbol	Fold change mature ovaries	Fold change early ovaries	Presence of CPE
FBgn0038893	CG6353		2.08	CPE
FBgn0047038	CG6463	2.24		
FBgn0035675	CG6610	2.35	2.42	
FBgn0037901	CG6744		1.82	CPE
FBgn0036030	CG6767	3.42	2.98	
FBgn0032400	CG6770	10.3	4.53	CPE
FBgn0036488	CG6878	2.43	3.03	
FBgn0038976	CG7048	1.56	2.02	
FBgn0038099	CG7091	2.13		CPE
FBgn0038575	CG7208	1.65		CPE
FBgn0038571	CG7215	4.1		
FBgn0031708	CG7382		1.86	
FBgn0036173	CG7394	2.47	2.22	CPE
FBgn0036762	CG7430	1.55	2.38	
FBgn0036745	CG7484	1.86	1.9	
FBgn0035807	CG7492	2.09		
FBgn0040833	CG7498	2.1	1.9	
FBgn0038108	CG7518	1.8		CPE
FBgn0037087	CG7519	2.7		CPE
FBgn0036728	CG7580	11.59	5.04	CPE
FBgn0036726	CG7603	1.56		
FBgn0032026	CG7627	1.56		
FBgn0040793	CG7630	6.97	3.48	
FBgn0033548	CG7637	10.7	7.29	CPE
FBgn0030883	CG7772	1.94		
FBgn0032020	CG7787	1.55	2.1	CPE
FBgn0039697	CG7834		2.23	
FBgn0039735	CG7911	13.69	6.11	
FBgn0038582	CG7988		1.68	
FBgn0037607	CG8036	1.6		
FBgn0034021	CG8180	1.53		
FBgn0034033	CG8204		1.92	
FBgn0035830	CG8209		1.65	CPE
FBgn0033351	CG8235	1.68	2.32	
FBgn0033906	CG8331	5.03	4.86	CPE
FBgn0034061	CG8386	2.82	3.32	
FBgn0031992	CG8498	1.71		
FBgn0035714	CG8549	1.82	2.02	
FBgn0029629	CG8636	8.74	4.34	
FBgn0030834	CG8675		2.36	
FBgn0031684	CG8680	3.58	2.6	
FBgn0031663	CG8891		1.83	
FBgn0040931	CG9034	1.9		
FBgn0030610	CG9065	1.58		
FBgn0031771	CG9140	1.7	2.08	CPE
FBgn0030718	CG9172	3.2	3.00	
FBgn0038183	CG9286	1.76	3.12	
FBgn0032511	CG9306	5.23	3.25	
FBgn0034576	CG9350	7.04	3.66	
FBgn0037063	CG9391	1.67		

FlyBaseID	Symbol	Fold change mature ovaries	Fold change early ovaries	Presence of CPE
FBgn0034618	CG9485		1.84	
FBgn0032101	CG9586		1.78	CPE
FBgn0040529	CG9603	7.84	4.23	
FBgn0031485	CG9643		2.34	CPE
FBgn0036667	CG9669	5.43	4.34	
FBgn0038149	CG9796	3.15		
FBgn0037637	CG9836	1.73		
FBgn0030734	CG9911	1.65		
FBgn0029504	CHES-1-like	1.55		
FBgn0043001	Chrac-16	1.66		
FBgn0024814	Clc	2.63	2.22	
FBgn0015622	Cnx99A	2.43		
FBgn0032833	CoIV	5.97	4.2	
FBgn0019624	CoVa	7.6	3.77	
FBgn0031830	CoVb	2.75		
FBgn0031066	CoVlb	2.16	2.52	
FBgn0040773	CoVIIc	6.4	3.42	
FBgn0263911	CoVIII	36.38	7.81	
FBgn0000355	Cp15	48.73	6.35	
FBgn0000356	Cp16	23.32		
FBgn0000357	Cp18	36.42	4.21	
FBgn0000358	Cp19	30.24	4.8	
FBgn0000359	Cp36	14.38		
FBgn0000360	Cp38	58.91		
FBgn0033598	Cpr47Eb		1.78	
FBgn0000392	cup	1.8		CPE
FBgn0000405	CycB	3.81		CPE
FBgn0039858	CycG	2.5		CPE
FBgn0032378	CycY	2.37		CPE
FBgn0015031	cype	5.88	4.37	
FBgn0000409	Cyt-c-p	8.56	3.44	
FBgn0000412	D1	1.94		
FBgn0010316	dap	7.1	3.08	CPE
FBgn0011761	dhd	33.64	8.3	
FBgn0004087	Dhfr	1.66	2.38	
FBgn0000447	Dhod	1.56	1.95	
FBgn0031601	Dim1		2.35	
FBgn0030276	Dlic	2.66		
FBgn0000479	dnc	1.55		
FBgn0024558	Dph5	3.85	3.63	
FBgn0266518	Dpit47	2.00	2.78	
FBgn0038145	Droj2	1.84		
FBgn0250837	dUTPase	3.93	4.26	CPE
FBgn0000618	e(y)2		1.98	
FBgn0011766	E2f	1.61		CPE
FBgn0027066	Eb1	2.06		
FBgn0263933	ebi		1.65	
FBgn0069242	eca	3.08	2.79	
FBgn0000556	Ef1alpha48D	34.19	7.36	
FBgn0028737	Ef1beta	10.87	4.52	

FlyBaseID	Symbol	Fold change mature ovaries	Fold change early ovaries	Presence of CPE
FBgn0029176	Ef1gamma	25.47	6.46	
FBgn0004926	eIF-2beta	3.37		
FBgn0001942	eIF-4a (CG9075-RC)	7.57	3.68	
FBgn0001942	eIF-4a (CG9075-RB)	1.52		
FBgn0015218	eIF-4E	3.33		
FBgn0023213	eIF4G	3.36	2.57	
FBgn0034967	eIF-5A	14.45	5.23	
FBgn0023212	Elongin-B		1.83	
FBgn0023211	Elongin-C	2.52	3.31	
FBgn0061515	endos		1.84	
FBgn0264693	ens	1.97		CPE
FBgn0037913	fabp	3.22		
FBgn0000644	Fcp3C	2.41		
FBgn0011768	Fdh	1.67		CPE
FBgn0041252	Femcoat	1.63		CPE
FBgn0039969	Fis1	1.55		CPE
FBgn0013269	FK506-bp1	3.78	2.32	
FBgn0263773	fok	4.32	3.24	
FBgn0001104	Galphai	2.71		CPE
FBgn0001091	Gapdh1	4.55	3.1	
FBgn0001092	Gapdh2	18.77		
FBgn0004868	Gdi	13.24	4.82	CPE
FBgn0033081	geminin	1.63		
FBgn0039580	Gfat2	2.09		
FBgn0011770	Gip	3.00		
FBgn0001120	gnu	3.7	4.7	CPE
FBgn0026430	Grip84	2.15		CPE
FBgn0001149	GstD1	6.03		CPE
FBgn0027590	GstE12	1.77	2.08	CPE
FBgn0035906	GstO2		2.35	CPE
FBgn0031117	GstT3	1.52	2.2	
FBgn0010391	Gtp-bp	2.1	2.03	
FBgn0014189	Hel25E	4.51		CPE
FBgn0051617	His1:CG31617	5.83		
FBgn0051618	His2A:CG31618	7.96		
FBgn0061209	His2B:CG17949		2.19	
FBgn0051613	His3:CG31613	1.51		
FBgn0051611	His4:CG31611	5.51	2.76	
FBgn0013981	His4r	2.09		CPE
FBgn0015393	hoip	12.16	4.92	
FBgn0035829	HP4	1.69		
FBgn0001218	Hsc70-3	2.5		
FBgn0001219	Hsc70-4	8.8		
FBgn0034491	Hsl		1.58	CPE
FBgn0001233	Hsp83	7.23		
FBgn0028429	I-2	1.5		
FBgn0024227	ial	1.94		CPE
FBgn0015247	lap2	1.79		
FBgn0025366	Ip259	3.16	3.26	
FBgn0036999	isoQC	1.87		

FlyBaseID	Symbol	Fold change mature ovaries	Fold change early ovaries	Presence of CPE
FBgn0040309	Jafrac1	5.14		
FBgn0040308	Jafrac2	1.56		
FBgn0001280	janA	1.84	2.72	
FBgn0250753	kra (CG2922-RC)	3.55		
FBgn0250753	kra (CG2922-RA)	2.61		
FBgn0040890	ksh	3.7	2.9	
FBgn0038476	kuk	3.63		
FBgn0001332	L	1.54		
FBgn0027291	l(1)G0156	1.84		
FBgn0028325	l(1)G0334	2.48		
FBgn0010612	l(2)06225	13.38	5.67	
FBgn0002021	l(2)37Bb	2.54		
FBgn0002031	l(2)37Cc	2.64		
FBgn0086447	l(2)37Cg		2.21	
FBgn0010741	l(3)01239	2.49	2.41	
FBgn0002283	l(3)73Ah		1.95	CPE
FBgn0002354	l(3)87Df	1.52		
FBgn0011455	l(3)neo18	2.22	2.51	
FBgn0011638	La	1.63		
FBgn0034877	levy	14.59	5.6	
FBgn0051092	LpR2	1.5		CPE
FBgn0261279	lqfR	1.67		
FBgn0033554	Lsm10		1.58	
FBgn0261068	Lsm7		2.18	
FBgn0037092	M6		1.78	CPE
FBgn0024332	Mcm3	1.89		
FBgn0262782	Mdh1	6.21	3.72	
FBgn0262559	Mdh2	6.97	5.29	
FBgn0004419	me31B	5.89	3.55	
FBgn0029155	Men-b		2.00	CPE
FBgn0034726	Mes4	1.91		
FBgn0035473	mge	1.61	2.39	
FBgn0031044	MKP-4	1.91	2.39	
FBgn0259482	Mob3		2.35	
FBgn0026409	Mpcp	5.3	4.27	
FBgn0033341	MrgBP	2.18	2.76	
FBgn0031231	mRpl10	2.83		
FBgn0038234	mRpl11	1.62		
FBgn0011787	mRpl12	3.64	3.83	
FBgn0032720	mRpl13	1.59	2.11	
FBgn0036990	mRpl15	1.7	2.44	
FBgn0036335	mRpl20	1.63		
FBgn0036853	mRpl21	2.00	2.18	
FBgn0035335	mRpl23	1.64		
FBgn0053002	mRpl27	2.57	3.00	
FBgn0031660	mRpl28	1.61	1.93	
FBgn0029718	mRpl30	2.59	2.84	
FBgn0039835	mRpl32	1.93	2.27	
FBgn0038923	mRpl35	1.81		
FBgn0030552	mRpl38	2.28	2.16	

FlyBaseID	Symbol	Fold change mature ovaries	Fold change early ovaries	Presence of CPE
FBgn0001995	mRpL4	1.53		
FBgn0037892	mRpL40	2.05	3.57	
FBgn0037330	mRpL44	3.43	4.05	
FBgn0028648	mRpL50	1.73	2.19	
FBgn0032053	mRpL51	1.92	2.67	
FBgn0050481	mRpL53	4.15	2.75	
FBgn0034579	mRpL54	1.8	2.39	
FBgn0038319	mRpL9	1.72		
FBgn0038307	mRpS10	1.88		
FBgn0038474	mRpS11	2.48	3.27	
FBgn0044030	mRpS14	2.85	2.62	
FBgn0033907	mRpS16	1.81	2.27	
FBgn0051450	mRpS18A	4.73		
FBgn0031639	mRpS2		1.98	
FBgn0044511	mRpS21	3.87		
FBgn0030572	mRpS25	1.81	2.47	
FBgn0038426	mRpS33	1.59		CPE
FBgn0035374	mRpS35	1.84	2.4	
FBgn0035209	msd1	1.99	2.09	
FBgn0011361	mtacp1	1.6	2.16	
FBgn0027786	Mtch	1.72		
FBgn0028479	Mtpalpha	2.09		
FBgn0010431	mtrm	4.32	3.56	CPE
FBgn0010438	mtSSB	5.17	3.31	
FBgn0261381	mtTFB1	1.7		
FBgn0005655	mus209		2.56	
FBgn0015268	Nap1	5.99	3.11	
FBgn0031020	Nat1	1.61		
FBgn0017567	ND23	3.47	2.4	
FBgn0019957	ND42	1.75	2.62	
FBgn0035046	NDUFA8	5.54	3.86	
FBgn0032725	Nedd8	1.78	2.41	
FBgn0261479	nero	2.92	2.57	
FBgn0029148	NHP2	2.02	2.57	CPE
FBgn0016685	Nlp	8.57	5.03	
FBgn0033224	Nop17l		1.55	CPE
FBgn0038964	Nop56	8.16	7.37	
FBgn0259937	Nop60B	2.44		CPE
FBgn0031381	Npc2a	5.29	3.82	
FBgn0034310	Nup75		1.69	CPE
FBgn0028411	Nxt1	5.11	3.21	
FBgn0014184	Oda	2.91		CPE
FBgn0027791	O-fut2	1.75	2.22	
FBgn0038870	Oga		3.24	
FBgn0041585	olf186-F	2.15		CPE
FBgn0015522	olf186-M	3.6	3.42	CPE
FBgn0003008	or	1.72	1.86	
FBgn0037110	ORMDL	1.94		
FBgn0016691	Oscp	10.35	7.29	
FBgn0003015	osk	7.4	3.98	CPE

FlyBaseID	Symbol	Fold change mature ovaries	Fold change early ovaries	Presence of CPE
FBgn0011336	OstStt3		2.24	CPE
FBgn0034255	Oxp		2.14	CPE
FBgn0033179	p47	1.79		
FBgn0265297	pAbp	2.67		
FBgn0005655	PCNA		2.56	
FBgn0036580	PDCD-5	3.08	3.21	
FBgn0014002	Pdi	5.54	3.01	CPE
FBgn0034058	Pex11	2.21	2.46	CPE
FBgn0032407	Pex19	1.65		
FBgn0023517	Pgam5	3.04	2.36	CPE
FBgn0016053	pgc	2.24		
FBgn0035438	PHGPx	2.75		
FBgn0086706	pix	1.6		CPE
FBgn0000826	png	1.74		
FBgn0032884	Pomp	3.72	3.22	
FBgn0004363	porin	1.6		
FBgn0050290	Ppcdc	2.13		
FBgn0086134	Prosalpha2	3.96	2.99	
FBgn0261394	Prosalpha3	4.37	3.36	
FBgn0004066	Prosalpha4	5.5	3.28	
FBgn0016697	Prosalpha5	2.57		
FBgn0250843	Prosalpha6	1.87		
FBgn0023175	Prosalpha7	5.31	3.83	CPE
FBgn0023174	Prosbeta2	2.00		
FBgn0026380	Prosbeta3	1.86		
FBgn0032596	Prosbeta4	2.19		
FBgn0250746	Prosbeta7	3.47	2.37	
FBgn0038519	Prx3		1.94	
FBgn0038570	Prx5	4.1		
FBgn0052412	QC	2.43	2.54	
FBgn0016700	Rab1	3.64	2.53	
FBgn0015794	Rab18	1.5		
FBgn0014009	Rab2	1.61		CPE
FBgn0014010	Rab5		2.09	CPE
FBgn0020618	Rack1	25.69	5.59	
FBgn0026777	Rad23	2.59	2.26	CPE
FBgn0025806	Rap2l	2.85	2.46	
FBgn0260944	Rbp1	7.35	4.47	
FBgn0260985	RfC4	1.75		
FBgn0021906	RFeSP	1.63	2.25	
FBgn0024196	robl	2.3		
FBgn0025638	Roc1a	4.66	3.68	
FBgn0039218	Rpb10	3.51	4.12	CPE
FBgn0038903	Rpl12	1.78		
FBgn0004855	RplI15		1.81	
FBgn0024733	RpL10	31.69	5.85	
FBgn0036213	RpL10Ab	57.06	10.2	
FBgn0013325	RpL11	35.32	9.36	
FBgn0034968	RpL12	31.68	6.79	
FBgn0011272	RpL13	56.69	13.91	

FlyBaseID	Symbol	Fold change mature ovaries	Fold change early ovaries	Presence of CPE
FBgn0037351	RpL13A	33.14	6.71	
FBgn0017579	RpL14	30.09	5.95	
FBgn0028697	RpL15	43.32		CPE
FBgn0035753	RpL18	4.7	3.32	CPE
FBgn0010409	RpL18A	4.00		
FBgn0002607	RpL19	56.08	10.29	
FBgn0032987	RpL21	22.67	6.05	
FBgn0015288	RpL22	4.72		CPE
FBgn0010078	RpL23	19.6	8.29	
FBgn0026372	RpL23A	30.62	8.74	CPE
FBgn0032518	RpL24	52.45	12.68	
FBgn0037899	RpL24-like	1.95		
FBgn0036825	RpL26	13.87	4.95	
FBgn0261606	RpL27A	54.51	13.72	
FBgn0016726	RpL29	2.53		
FBgn0020910	RpL3	11.17	6.69	
FBgn0086710	RpL30	18.27	7.51	CPE
FBgn0025286	RpL31	46.64	9.67	CPE
FBgn0039406	RpL34a	13.02	7.65	
FBgn0029785	RpL35	56.99	12.18	
FBgn0037328	RpL35A		1.74	
FBgn0031980	RpL36A	58.51	14.02	CPE
FBgn0261608	RpL37A	50.94	11.7	CPE
FBgn0030616	RpL37a	47.45	10.89	CPE
FBgn0040007	RpL38	14.37	7.93	
FBgn0023170	RpL39	23.69	10.93	
FBgn0003279	RpL4	35.2	10.08	CPE
FBgn0003941	RpL40	11.11	4.54	
FBgn0066084	RpL41	52.56	14.94	
FBgn0064225	RpL5	67.58	8.62	
FBgn0039857	RpL6	8.96	4.01	
FBgn0005593	RpL7	41.94	13.3	CPE
FBgn0014026	RpL7A	9.64	4.89	CPE
FBgn0261602	RpL8	31.68	6.79	
FBgn0015756	RpL9	42.01	8.36	
FBgn0000100	RpLP0	56.27	14.61	
FBgn0033485	RpLP0-like		1.64	CPE
FBgn0002593	RpLP1	59.6	18.95	
FBgn0003274	RpLP2	93.61	13.57	
FBgn0028694	Rpn11	1.87		
FBgn0028692	Rpn2	1.59		
FBgn0261593	RpS10b	51.88	8.15	
FBgn0033699	RpS11	53.27	11.1	
FBgn0260441	RpS12	25.66	5.29	
FBgn0010265	RpS13	35.38	10.64	
FBgn0004403	RpS14a	51.58	16.45	
FBgn0004404	RpS14b	2.97	3.09	
FBgn0034138	RpS15	27.76	10.17	CPE
FBgn0034743	RpS16	60.38	13.27	
FBgn0010411	RpS18	34.23	12.28	

FlyBaseID	Symbol	Fold change mature ovaries	Fold change early ovaries	Presence of CPE
FBgn0010412	RpS19a	28.79	9.64	
FBgn0004867	RpS2	71.48	7.39	
FBgn0019936	RpS20	17.09	12.59	
FBgn0015521	RpS21 (CG2986-RC)	27.97	9.32	
FBgn0015521	RpS21 (CG2986-RD)	2.02	1.95	
FBgn0033912	RpS23	34.99	12.49	
FBgn0261596	RpS24	21.06	10.23	
FBgn0261597	RpS26	26.76	8.16	
FBgn0039300	RpS27	19.03	6.07	
FBgn0003942	RpS27A	60.62	12.29	
FBgn0030136	RpS28b	21.46	5.85	
FBgn0261599	RpS29	14.42	4.00	
FBgn0002622	RpS3	14.01	7.07	
FBgn0038834	RpS30	43.52	9.72	
FBgn0017545	RpS3A	33.59	5.9	CPE
FBgn0011284	RpS4	63.24	10.53	
FBgn0002590	RpS5a	7.44	4.15	CPE
FBgn0038277	RpS5b	27.85	8.2	
FBgn0261592	RpS6	23.98	5.61	
FBgn0039757	RpS7	41.01	10.4	
FBgn0039713	RpS8 (CG7808-RB)	53.96	13.99	
FBgn0039713	RpS8 (CG7808-RC)	26.67	6.25	
FBgn0010408	RpS9	66.64	15.03	
FBgn0053113	Rtnl1	1.66		CPE
FBgn0038947	Sar1	1.61		CPE
FBgn0035471	Sc2	1.7		
FBgn0265298	SC35	2.97		
FBgn0041094	scyl	3.9		CPE
FBgn0014028	SdhB	2.71	1.95	
FBgn0037873	SdhC	2.74	2.11	
FBgn0037912	sea	3.22		
FBgn0010638	Sec61beta	3.09		
FBgn0031049	Sec61gamma	5.52		
FBgn0003360	sesB	17.27	7.62	
FBgn0030486	Set2	1.67		
FBgn0041186	Slbp	2.14	1.91	CPE
FBgn0262601	SmB	2.52		
FBgn0261790	SmE	1.56		
FBgn0000426	SmF	5.89	4.25	
FBgn0261791	SmG	7.77	4.31	
FBgn0036641	Smn	2.00		
FBgn0264922	smt3	8.28	3.24	CPE
FBgn0265192	Snf (CG6393-RD)	2.16		
FBgn0265192	Snf (CG6393-RC)	1.95		
FBgn0011715	Snr1		1.85	
FBgn0003462	Sod	1.62		
FBgn0033631	Sod3	2.93		
FBgn0040623	Spase12	1.53		
FBgn0030306	Spase25	3.14	2.76	
FBgn0087021	Spc25	1.51		

FlyBaseID	Symbol	Fold change mature ovaries	Fold change early ovaries	Presence of CPE
FBgn0037723	SpdS	3.71	4.47	
FBgn0028683	spt4	1.77		
FBgn0014032	Sptr	1.55	1.9	
FBgn0263396	sqd	3.75		CPE
FBgn0038808	Srp14	4.52	5.04	CPE
FBgn0035947	Srp68	1.56		
FBgn0015299	Ssb-c31a	1.76	2.38	
FBgn0011481	Ssdp	2.27		
FBgn0011016	SsRbeta	3.96		
FBgn0003517	sta (CG14792-RA)	8.8	4.56	
FBgn0003517	sta (CG14792-RD)	6.55	3.88	
FBgn0266521	stai		2.21	
FBgn0003525	stg	4.26		
FBgn0046692	Stlk	1.59		
FBgn0004465	Su(P)	1.61	1.86	CPE
FBgn0003612	Su(var)2-10	1.87		CPE
FBgn0021795	Tapdelta	4.36	3.96	
FBgn0027329	Tcp-1zeta	1.67		
FBgn0037874	Tctp	20.87	5.96	
FBgn0261014	TER94	2.12		
FBgn0027360	Tim10	11.38	5.31	
FBgn0263977	Tim17b	3.29	3.1	
FBgn0030480	Tim9a	2.19	2.53	
FBgn0036928	Tom20	5.27		CPE
FBgn0033357	Tom7	4.83	5.28	
FBgn0015834	Trip1	2.38	2.31	
FBgn0024921	Trn	4.51	2.87	CPE
FBgn0260861	Trs23	1.79	2.27	
FBgn0011726	tsr	5.3	2.99	
FBgn0033378	tsu	2.72		
FBgn0250874	ttm50	1.59		
FBgn0015320	UbcD2	1.67		
FBgn0015321	UbcD4	1.53		
FBgn0029996	Ubc-E2H	1.58		
FBgn0022224	ubl		2.82	CPE
FBgn0027526	Ublcp1	1.53		CPE
FBgn0010288	Uch	1.75		
FBgn0035978	UGP	1.54		CPE
FBgn0014075	Ugt		1.83	CPE
FBgn0045800	Uhg1		2.97	
FBgn0259936	Uhg3	2.02		
FBgn0086691	UK114	7.19	4.22	
FBgn0025117	und	2.69	2.73	
FBgn0033452	Vamp7	2.81		
FBgn0026753	Vha13	12.36	5.3	
FBgn0262512	Vha14-1	3.96	3.31	
FBgn0028663	VhaM9.7-b	2.02	2.5	CPE
FBgn0003979	Vm26Aa	32.19	6.56	
FBgn0003980	Vm26Ab	15.17	4.11	
FBgn0086266	Vm26Ac		1.88	

FlyBaseID	Symbol	Fold change mature ovaries	Fold change early ovaries	Presence of CPE
FBgn0014076	Vm32E	4.83	2.94	
FBgn0003983	Vm34Ca	39.69	6.98	
FBgn0260987	vtd	3.06		
FBgn0010516	wal	3.1	2.59	
FBgn0041709	yellow-g	9.23		
FBgn0043842	Yeti	2.27		
FBgn0004045	Yp1	10.59		
FBgn0022959	yps	11.33	4.82	CPE
FBgn0040512	zetaCOP	1.98		